Soybean cropland as foraging source for the European honey bee and predictor of honey content

THESIS

Submitted in Fulfillment of the Requirements for Undergraduate Research Distinction

By

Luke Hearon, Chia-Hua Lin, and Reed Johnson

The Ohio State University

2019

Abstract

Soybean (Glycine max) farming contributes a large portion of Ohio's annual crop revenue and accounts for a similarly large portion of Ohio's cropland. Though *G. max* is self-pollinated, and thus not dependent on insect pollinators, it has been found that insect pollinators will visit *G. max* flowers nonetheless, and in doing so cause an increase in soybean yield. Conventionally, soybean has not been considered a major source of nectar for the summertime foraging of honey bees (Apis mellifera). Over the last decade, it has become increasingly obvious that, to an uncertain degree, *G. max* bloom does contribute to honey production. We seek to determine the the extent to which *G. max* contributes to honey in Ohio. Samples of honey from hives across the state were collected from participating beekeepers. The pollen grains were extracted from each sample, counted via light microscopy, and classified into their respective taxa. A nearly-pure soybean honey was created to determine quantity of *G. max* pollen grains per gram. The ratio was found to be 7,007 grains of soybean pollen per gram of soybean honey. This ratio was then applied to the *G. max* pollen count of each honey sample to infer what proportion by mass of that honey is derived from soybean. Geographic information system (GIS) analysis was used to determine the presence of soybean cropland within foraging range of the hives at the time samples were taken. A high proportion (70%) of samples were derived in part from soybean, most of these at less than 5% by mass. Nine samples, however, are comprised of 10% or greater soybean honey by mass, the greatest of which being 44% soybean honey. When correlated with the presence of soybean as determined in GIS analysis, a weak positive relationship may be established between soybean presence and soybean composition of honeys. The implications of foraging on *G. max* by honey bees are twofold: to the crop grower and to the beekeeper. A clearer understanding of soybean pollination will allow farmers to make more informed choices

about how and when to apply pesticides and allow beekeepers to position their hives to best maximize productivity and avoid potentially injurious pesticides.

Background

In 2018, soybean (*Glycine max*) accounted for 5.09 million acres of Ohio cropland (Turner and Morris, 2018). Many studies have shown that honey bees (*Apis mellifera*) will forage upon, and in doing so pollinate, soybean (Milfont et al., 2013; Monasterolo et al., 2015; Vanderlinden, 1981; Chiari et. al., 2015). Pollination by honey bees has been shown to increase the yield of the soybean significantly. Yield increased 11.75% when exposed to honey bees (Milfont et al., 2013).

Analysis of pollen found in honey samples provides a proxy by which foraging behavior of honey bees can be examined. However, because there is extreme variation in pollen output between species of flowers, it is worthless to examine relative frequencies of pollen types in honey. The counted pollen grains must be adjusted with a pollen coefficient—the number of pollen grains found in one gram of honey purely from that flower (Bryant and Jones, 2001). Only after this adjustment can a proportion of honey attributable to that flower can be determined. No satisfactory coefficient has been created for soybean honey.

Because many experiments involving pollination of soybean by honey bees employ low- or no-choice tests (Milfont et al., 2013 and Chiari et. al., 2015) or examine effects upon soybean rather than upon honey bees (Monasterolo et al., 2015), understanding of the foraging habits of honey bees is still incomplete.

Objectives

The objectives of this study were threefold: to determine the pollen coefficient of soybean, to examine foraging habits of honey bees upon soybean in Ohio, and to determine the extent to which soybean contributes to summer honeys in Ohio.

Methods

Pollen Analysis

45 honey samples and their respective coordinates were contributed by volunteering beekeepers across Ohio. Pollen from these samples was extracted by dilution with ethanol and centrifugation. Pollen grains were counted and classified via light microscopy.

A pure soybean honey was made by placing two 5-frame hives with new foundation in the midst of a large soybean field and gathering the resultant honey. The pollen coefficient was calculated from this pure soybean honey and used to determine the proportion by mass of soybean honey in each sample.

GIS Analysis

The landscape around each hive was analyzed in ArcGIS (ESRI) using the Ohio crop data layer from the USDA's CropScape project. Each raster cell (a 30m by 30m patch) identified as soybean was weighted for its distance from the hive in question according to the patch visitation probability from Sponsler (2016). The values of all cells within 3km of the hive were summed, creating the single value of "soybean presence" by which to rank the sites.

Results

The pollen coefficient of soybean honey was found to be 7,007 grains of soybean pollen per gram of pure soybean honey. The soybean honey created was nearly pure, but because a few non-soybean pollens were observed, this pollen coefficient is, to some degree, an approximation.

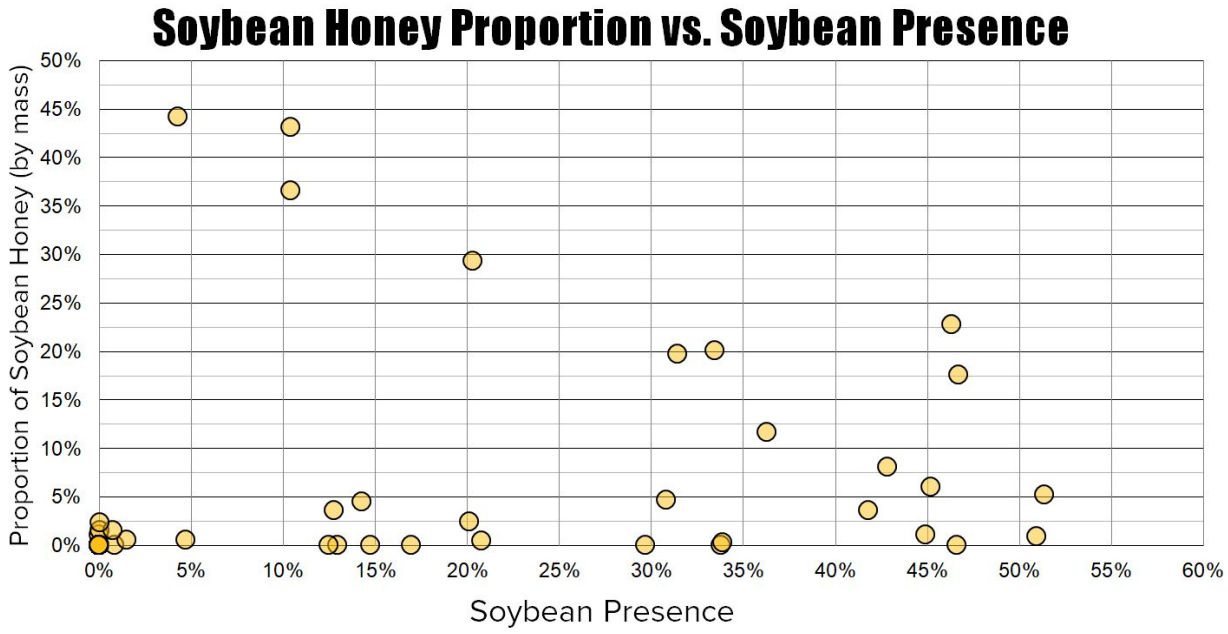

Figure 1. Proportion by mass of soybean honey in honey samples compared to presence of soybean around hive.

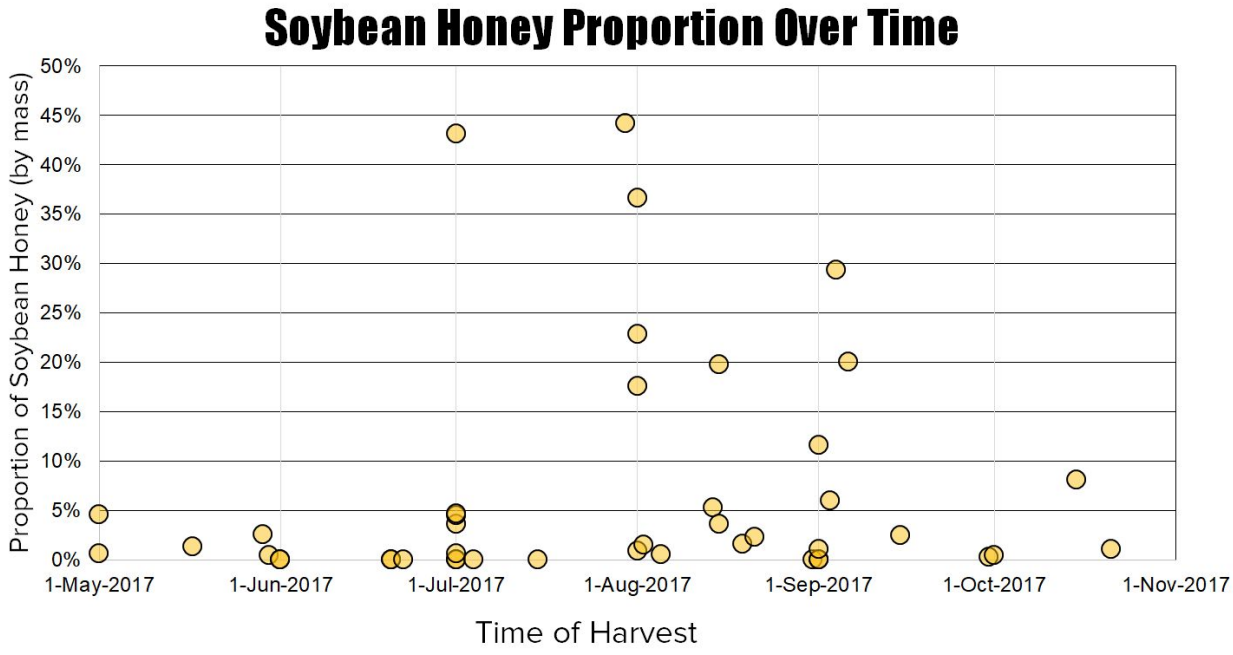

Figure 2. Proportion by mass of soybean in each honey sample with respect to time of harvest. Soybean bloom begins in early July in most of Ohio.

The proportion of soybean honey in samples was seen to be below 5% before soybean bloom. In July, when most soybean begins to bloom in Ohio, the proportion of soybean honey drastically increases. Honeys harvested later in soybean bloom showed a lower proportion of soybean honey.

Over 70% of samples contained some amount of soybean honey, but only 9 samples are comprised of more than 10% soybean honey by mass. The two samples containing the most soybean honey were comprised of over 40% soybean honey. When correlating proportion of soybean honey to soybean presence in the surrounding landscape, there exists a weak, but statistically significant ($p < 0.05$) Spearman's rank-order correlation of $\rho = 0.295$.

Conclusions

The relationship between soybean presence and its proportion in honey is monotonic, positive, and weak. This may indicate a low preference by honey bees for soybean as forage. While not always a major component, soybean nectar contributes to the majority of summer honeys of Ohio. Beekeepers in Ohio should expect their bees to be foraging upon soybean when it is in bloom, particularly in July. In contrast to low levels in most samples, a few hives showed heavy foraging upon soybean with nearly half of the honey sampled from two hives being made from soybean honey.

Though soybean pollen was found in samples harvested before soybean bloom, this is not unexpected. We suspect that this is due to residual soybean pollen in the honeycomb from years past.

The four samples which showed the greatest proportion of soybean honey by mass were found in areas of low soybean presence. This merits further investigation.

Acknowledgements

I would like to thank the following for their invaluable assistance to this project:
The Ohio State Beekeeper's Association for funding this research effort.
The beekeepers of the Ohio State Beekeeper's Association, who graciously supplied the honey samples essential to this study and multiple others.
Joshua Sadvari, who helped me to create a smooth and efficient workflow for the GIS analysis protocol.

References

Bryant, V. M., & Jones, G. D. 2001. *The r-values of honey: Pollen coefficients.* Palynology: 25(1): 11-28.

Milfont, de O., E.E.M. Rocha, A.O.N. Lima, and B.M. Freitas. 2013. *Higher soybean production using honeybee and wild pollinators, a sustainable alternative to pesticides and autopollination.* Environmental Chemistry Letters 11(4): 335-341.

Monasterolo M., M.L. Musicante, G.R. Valladares, and A. Salvo. 2015. *Soybean crops may benefit from forest pollinators*. Agriculture, Ecosystems & Environment 202: 217-222

Sponsler, D.B. 2016. *Honey bee landscape ecology: foraging, toxic exposure, and apicultural outcomes*. Retrieved from: The Ohio State University Libraries.

Turner, C., B. Morris. 2018. *Ohio Agricultural Statistics 2017-2018 Annual Bulletin*. United States Department of Agriculture.

Vanderlinden, J.O. 1981. *Soybean honey production in Iowa.* American bee journal 121(10): 723.

Chiari, W. C., Toledo, V. D. A. A. D., Ruvolo-Takasusuki, M. C. C., Oliveira, A. J. B. D., Sakaguti, E. S., Attencia, V. M., ... & Mitsui, M. H. (2005). Pollination of soybean (Glycine max L. Merril) by honeybees (Apis mellifera L.). Brazilian Archives of Biology and Technology, 48(1), 31-36.